

Louisiana Office of Public Health Laboratories	
Test Name	Middle East Respiratory Syndrome Coronavirus Emergency Use Authorization Procedure
PHL Location	Office of Public Health Laboratory Baton Rouge
CPT Code	87798
Synonyms	MERS CoV, Middle East Respiratory Syndrome Coronavirus, Novel Coronavirus 2012, NCV-2012
Brief Description of Test	Prior authorization required. Contact Infectious Disease Epidemiology at 800-256-2748. Real-time (TaqMan®) RT-PCR (rRT-PCR) assay for detection of the Middle East Respiratory Syndrome Coronavirus (MERS-CoV), previously known as Novel Coronavirus 2012 or NCV-2012
Possible Results	MERS-CoV RNA not detected by rRT-PCR Inconclusive for MERS-CoV RNA by rRT-PCR. An inconclusive result may occur in the case of an inadequate specimen. MERS-CoV RNA detected by rRT-PCR. Confirmatory testing required. Specimen will be referred to CDC for further analysis. NCV-2012 rRT-PCR testing was equivocal. Additional analysis may be conducted by CDC.
Reference Range	Not Detected
Specimen Type	Nasopharyngeal and/or Oropharyngeal swabs Sputum Lower respiratory tract aspirates/washes Serum Stool (Not acceptable with our extraction method but sample is acceptable to forward to CDC)
Specimen Container(s):	Sterile containers and/or Viral Transport Media Tubes
Minimum volume accepted:	See collection instructions
Collection Instructions	A. Lower respiratory tract Bronchoalveolar lavage, tracheal aspirate, pleural fluid Collect 2-3 mL into a sterile, leak-proof, screw-cap sputum collection cup or sterile dry container. Refrigerate specimen at 2-8°C up to 72 hours; if exceeding 72 hours, freeze at -70°C and ship on dry ice. Sputum

Have the patient rinse the mouth with water and then expectorate deep cough sputum directly into a sterile, leak-proof, screw-cap sputum collection cup or sterile dry container. Refrigerate specimen at 2-8°C up to 72 hours; if exceeding 72 hours, freeze at -70°C and ship on dry ice.

B. Upper respiratory tract

Nasopharyngeal AND oropharyngeal swabs (NP/OP swabs)

Use only synthetic fiber swabs with plastic shafts. Do not use calcium alginate swabs or swabs with wooden shafts, as they may contain substances that inactivate some viruses and inhibit PCR testing. Place swabs immediately into sterile tubes containing 2-3 ml of viral transport media. **NP/OP specimens can be combined, placing both swabs in the same vial.** Refrigerate specimen at 2-8°C up to 72 hours; if exceeding 72 hours, freeze at -70°C and ship on dry ice.

Nasopharyngeal swabs - Insert a swab into the nostril parallel to the palate. Leave the swab in place for a few seconds to absorb secretions. Swab both nasopharyngeal areas.

Oropharyngeal swabs - Swab the posterior pharynx, avoiding the tonsils and tongue.

Nasopharyngeal wash/aspirate or nasal aspirates

Collect 2-3 mL into a sterile, leak-proof, screw-cap sputum collection cup or sterile dry container. Refrigerate specimen at 2-8°C up to 72 hours; if exceeding 72 hours, freeze at -70°C and ship on dry ice.

II. Blood Components

Serum (for serologic testing at CDC)

For serum antibody testing: Serum specimens should be collected during the acute stage of the disease, preferably during the first week after onset of illness, and again during convalescence, ≥ 3 weeks after the acute sample was collected. However, since we do not want to delay detection at this time, a single serum sample collected 14 or more days after symptom onset may be beneficial. Serologic testing is currently available at CDC upon request and approval. Please be aware that the MERS-CoV serologic test is for research/surveillance purposes and not for diagnostic purposes - it is a tool developed in response to the MERS-CoV outbreak. Contact CDC's Emergency Operations Center (EOC) (770-488-7100) for consultation and approval if serologic testing is being considered.

Serum (for rRT-PCR testing)

For rRT-PCR testing (i.e., detection of the virus and not antibodies), a single serum specimen collected optimally during the first week after symptom onset, preferably within 3-4 days, after symptom onset, may also be beneficial.

Note: These time frames are based on SARS-CoV studies. The kinetics of MERS-CoV are not well understood and may differ from SARS-CoV. Once additional data become available, these recommendations will be updated as needed.

Children and adults. Collect 1 tube (5-10 mL) of whole blood in a serum separator tube. Allow the blood to clot, centrifuge briefly,

	<p>and separate sera into sterile tube container. The minimum amount of serum required for testing is 200 µL. Refrigerate the specimen at 2-8°C and ship on ice- pack; freezing and shipment on dry ice is permissible.</p> <p>Infants. A minimum of 1 mL of whole blood is needed for testing of pediatric patients. If possible, collect 1 mL in an EDTA tube and in a serum separator tube. If only 1 mL can be obtained, use a serum separator tube.</p> <p>EDTA blood (plasma) Collect 1 tube (10 mL) of heparinized (green-top) or EDTA (purple-top) blood. Refrigerate specimen at 2-8°C and ship on ice-pack; do not freeze.</p> <p>III. Stool Collect 2-5 grams of stool specimen (formed or liquid) in sterile, leak-proof, screw-cap sputum collection cup or sterile dry container. Refrigerate specimen at 2-8°C up to 72 hours; if exceeding 72 hours, freeze at -70°C and ship on dry ice.</p>
Storage and Transport Instructions	<p>For short periods (≤ 72 hours), most specimens should be held at 2-8°C rather than frozen. For delays exceeding 72 hours, freeze specimens at -70°C as soon as possible after collection (with exceptions noted above).</p> <p>Specimens from suspected MERS cases must be packaged, shipped, and transported according to the current edition of the International Air Transport Association (IATA) Dangerous Goods Regulations.² Specimens should be stored and shipped at the temperatures indicated above. If samples are unable to be shipped within 72 hours of collection, they should be stored at -70°C and shipped on dry ice. When shipping frozen specimen from long distances or from international locations, it is best to use a combination of dry ice and frozen gel ice-packs. The gel ice-packs will remain frozen for a day or two after the dry ice has dissipated.</p> <p>All specimens must be pre-packed to prevent breakage and spillage. Specimen containers should be sealed with Parafilm® and placed in ziplock bags. Place enough absorbent material to absorb the entire contents of the Secondary Container (containing Primary Container) and separate the Primary Containers (containing specimen) to prevent breakage. Send specimens with cold packs or other refrigerant blocks that are self-contained, not actual wet ice. This prevents leaking and the appearance of a spill. When large numbers of specimens are being shipped, they should be organized in a sequential manner in boxes with separate compartments for each specimen.</p>
Causes for Rejection	<p>Samples that do not meet time, temperature or documentation criteria. Samples that arrive on expired transport media. Samples from unacceptable specimen sources.</p>
Limitations of the Procedure	<p>Interpretation of rRT-PCR test results must account for the possibility of false-negative and false-positive results. False-negative results can arise from poor sample collection or</p>

	<p>degradation of the viral RNA during shipping or storage. Application of appropriate assay controls that identify poor-quality samples can help avoid most false-negative results. A more difficult problem is the apparently low titer of virus shed in specimens collected early in illness, which may make it difficult to confirm a diagnosis. The most common cause of false-positive results is contamination with previously amplified DNA. The use of rRT-PCR helps mitigate this problem by operating as a contained system. A more difficult problem is the cross-contamination that can occur between specimens during collection, shipping, and aliquoting in the laboratory. Liberal use of negative control samples in each assay and a well-designed plan for confirmatory testing can help ensure that laboratory contamination is detected and that false positive test results are not reported.</p> <p>The diagnosis of MERS-CoV infection must be made based on history, signs, symptoms, exposure likelihood, and other laboratory evidence, in addition to the detection of MERS-CoV RNA.</p> <p>Negative NCV-2012 rRT-PCR Assay results do not preclude MERS-CoV infection and should not be used as the sole basis for patient management decisions.</p> <p>The NCV-2012 rRT-PCR Assay is intended for use by trained laboratory personnel who are proficient in performing real-time RT-PCR assays. The level of MERS-CoV that would be present in respiratory specimens, sera or stool from individuals with early systemic infection is unknown. Due to the difficulty in obtaining positive clinical specimens, only limited evaluation of the NCV-2012 rRT-PCR Assay has been made with specimens from individuals with MERS-CoV infection.</p> <p>The NCV-2012 rRT-PCR Assay is only for use under the Food and Drug Administration's Emergency Use Authorization. Use within the United States is limited to qualified laboratories with training, facilities and equipment appropriate for specimen handling, testing and interpretation of the results of this real-time RT-PCR assay.</p>
Interfering Substances	N/A
References	Novel Coronavirus 2012 Real-Time RT-PCR Assay package insert EUA

Additional Information	Requires prior authorization from Infectious Disease Epidemiology for investigation.
Release Date	03/15/2016
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